ZooKeys 49: 1–11 (2010)
doi: 10.3897/zookeys.49.402
www.pensoftonline.net/zookeys

RESEARCH ARTICLE

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Two new species of nematode (Oxyurida, Hystrignathidae) parasites of *Passalus interstitialis* Escholtz, 1829 (Coleoptera, Passalidae) from Cuba and a new locality for *Longior similis* Morffe, García & Ventosa, 2009

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Academic editor: *David Gibson* | Received 22 April 2010 | Accepted 26 January 2010 | Published 18 June 2010

urn:lsid:zoobank.org:pub:29E83894-3732-4169-8A18-59FA3EBBABCE

Citation: Morffe J, García N (2010) Two new species of nematode (Oxyurida, Hystrignathidae) parasites of *Passalus interstitialis* Escholtz, 1829 (Coleoptera, Passalidae) from Cuba and a new locality for *Longior similis* Morffe, García & Ventosa, 2009. ZooKeys 49: 1–11. doi: 10.3897/zookeys.49.402

Abstract

Two new species of hystrignathids (Oxyurida: Hystrignathidae) are described as parasites of *Passalus interstitialis* Escholtz, 1829 from Cuba. *Hystrignathus splendidus* **sp. n.** differs from *H. inflatus* Travassos & Kloss, 1957 by having the eggs ridged, a stouter body and a shorter tail, and from *H. tarda* (Artigas, 1928) by its eggs being ridged and larger. *Lepidonema magnum* **sp. n.** can be differentiated from *L. brasiliensis* Travassos & Kloss, 1957 by the extension of the lateral alae, length of the first cephalic annule and the stouter body. It differs from *L. teresae* García, Ventosa & Morffe, 2009 by the esophagus and tail being comparatively shorter. *L. bifurcata* Cobb, 1898 differs from the new species by having the tail tip bifurcated. *L. caracae* Kloss, 1962 has more extended lateral alae and a shorter esophagus. Keys to the Cuban species of *Hystrignathus* and *Lepidonema* are given. *Longior similis* Morffe, García & Ventosa, 2009 is recorded from El Pan de Matanzas, Matanzas Province, Cuba.

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Keywords
Nematoda, Hystrignathidae, *Hystrignathus*, *Lepidonema*, *Longior*, Passalidae, *Passalus*, Cuba, parasites, new species

Introduction

The family Passalidae consists of a group of saproxylophagous and pan-tropical beetles with about 650 known species (Reyes-Castillo 2000). Due to their feeding habits passalids are frequently parasitized by monoxenous nematodes belonging to the family Hystrignathidae.

Two species of Passalidae have been recorded from Cuba: *Passalus pertyi* Kaup, 1869 (endemic) and *P. interstitialis* Escholtz, 1829. The latter is also distributed from Mexico to Argentina, including Jamaica (Peck 2005). However, only in Cuba has it been the object of parasitological studies. At present, six species, belonging to the genera *Artigasia*, *Glaber*, *Hystrignathus*, *Longior* and *Salesia*, are recorded as parasitizing *P. interstitialis* in Cuba (Coy 1990, Coy et al. 1993, García & Coy 1995a, b, García et al. 2009a, Morffe et al. 2009).

In this paper two new species of hystrignathid parasites of *P. interstitialis* are described and a new locality for *Longior similis* Morffe, García and Ventosa, 2009 is recorded from the same host.

Materials and methods

Nine specimens of *Passalus interstitialis* Escholtz, 1829 (eight from Escaleras de Jaruco, La Habana Province and one from El Pan de Matanzas, Matanzas Province) were collected by hand from rotting logs. The beetles were maintained alive in jars with moistened wood chips as food until dissection. They were killed in a killing jar with ethyl ether or acetone and immediately dissected according to Morffe et al. (2009). Nematodes extracted from their guts were killed with hot water (60–70°C) and fixed in 70% ethanol.

Specimens were clear-mounted in glycerine on glass slides and the edges of coverslips sealed with nail polish (Jex et al. 2004). Measurements were taken according to Travassos and Kloss (1958) and are given in millimeters. Variables are showed as range followed by median plus standard deviation in parentheses. De Man's ratios a, b, c and V% were calculated.

Parasites were photographed with an AxioCam digital camera attached to a Carl Zeiss compound microscope. Line drawings were made with CorelDRAW X3 and Adobe Photoshop CS2 using micrographs as templates. Scales of all plates are given in millimeters.

The type specimens are deposited in the Colección Helmintológica from the Colecciones Zoológicas del Instituto de Ecología y Sistemática, Havana, Cuba (CZACC).

Systematics

Genus *Hystrignathus* Leidy, 1850

Key to the species of *Hystrignathus* Leidy, 1850 from Cuba

Note: in the key we omit two species of Cuban hystrignathids formerly placed in *Hystrignathus*, which will be published in the future as new combinations.

1. First cephalic annule markedly inflated; spines ending at level of procorpus... ..... ***H. splendidus* Morffe & García, sp. n.**
– First cephalic annule not markedly inflated; spines surpassing the end of procorpus..... ***H. rosario* García, Ventosa & Morffe, 2009**

***Hystrignathus splendidus* sp. n.**
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Figs 1 A–I, 2 A–E

Type material. ♀ holotype, Cuba, La Habana Province, Jaruco, Escaleras de Jaruco; in *Passalus interstitialis*; 16.III.2008; E. Fonseca, J. Morffe & F. Alvarez coll.; CZACC 11.4530. 2 ♀♀ paratypes, same data as holotype, CZACC 11.4531–11.4532.

Measurements. Holotype (female) a = 15.28, b = 5.85, c = 7.24, V% = 54.55, total length = 2.750, maximum body width = 0.180, stoma length = 0.063, procorpus length = 0.360, isthmus length = 0.038, diameter of basal bulb = 0.078, total length of esophagus = 0.470, nerve ring to anterior end = 0.230, excretory pore to anterior end = 0.720, vulva to posterior end = 1.250, anus to posterior end = 0.380, eggs = 0.100–0.105×0.045–0.050 (0.104 ± 0.003×0.048 ± 0.002).

Paratypes (females) (n = 2): a = 14.71–15.89 (15.30 ± 0.84), b = 5.30–5.56 (5.43 ± 0.18), c = 5.71–6.41 (6.06 ± 0.50), V% = 53.93–54.00 (53.97 ± 0.05), total length = 2.225–2.500 (2.363 ± 0.194), maximum body width = 0.140–0.170 (0.155 ± 0.021), stoma length = 0.053 (0.053), procorpus length = 0.310–0.340 (0.325 ± 0.021), isthmus length = 0.035 (0.035), diameter of basal bulb = 0.068–0.078 (0.073 ± 0.007), total length of esophagus = 0.420–0.450 (0.435 ± 0.021), nerve ring to anterior end = 0.195–0.250 (0.223 ± 0.039), excretory pore to anterior end = 0.560–0.720 (0.640 ± 0.113), vulva to posterior end = 1.025–1.150 (1.088 ± 0.088), anus to posterior end = 0.390 (0.390), eggs = 0.098–0.108×0.043–0.053 (0.104 ± 0.003×0.050 ± 0.004).

Description. Female body comparatively stout. Cuticle markedly annulated in spiny region and less in rest of body. Cervical cuticle with opposite rows of small spines, from end of first cephalic annule at or near end of procorpus. Spines arranged initially in 16 rows increasing to about 24 rows where spines cease. Longitudinal sub-cuticular striae present. Lateral alae extending from end of spiny region

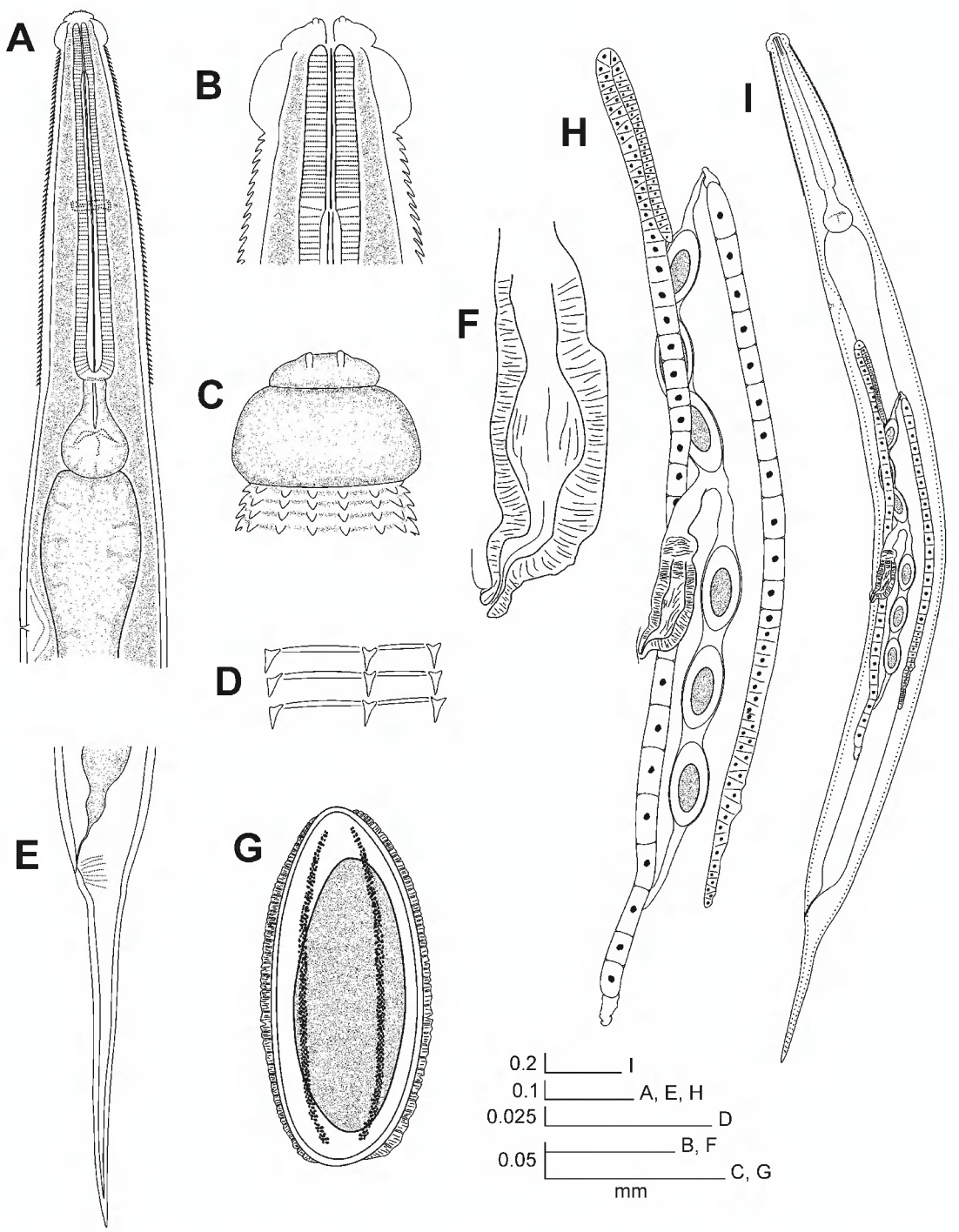

Figure 1. *Hystrignathus splendidus* sp. n. female. **A** Esophageal region, lateral view. **B** Cephalic end, internal view **C** Cephalic end, external view **D** Spines at level of the end of procorpus **E** Tail, lateral view **F** Vulva, lateral view **G** Egg. **H** Genital tract **I** Habitus, lateral view.

to level of vulva. Head bears eight papillae arranged in pairs. First cephalic annule long and inflated, about three head-lengths long and set-off from head by single groove. Stoma long, extending for about first two annule lengths and surrounded by an esophageal collar. Esophagus consisting of muscular procorpus with base slightly clavate and set-off from the isthmus. Basal bulb sub-pyriform with valve plate well developed. Intestine simple, sub-rectilinear and with its fore region inflated. Rectum short and anus not prominent. Nerve ring encircles procorpus at about its midpoint. Excretory pore situated at about one body-width posterior to basal bulb. Vulva a median transverse slit slightly displaced towards posterior half of body; lips not very prominent. Vagina muscular, forwardly directed. Genital tract didelphic-amphidelphic. Anterior ovary reflexed at about half of body-width posterior to excretory pore, with distal flexure about a body-width long. Posterior ovary

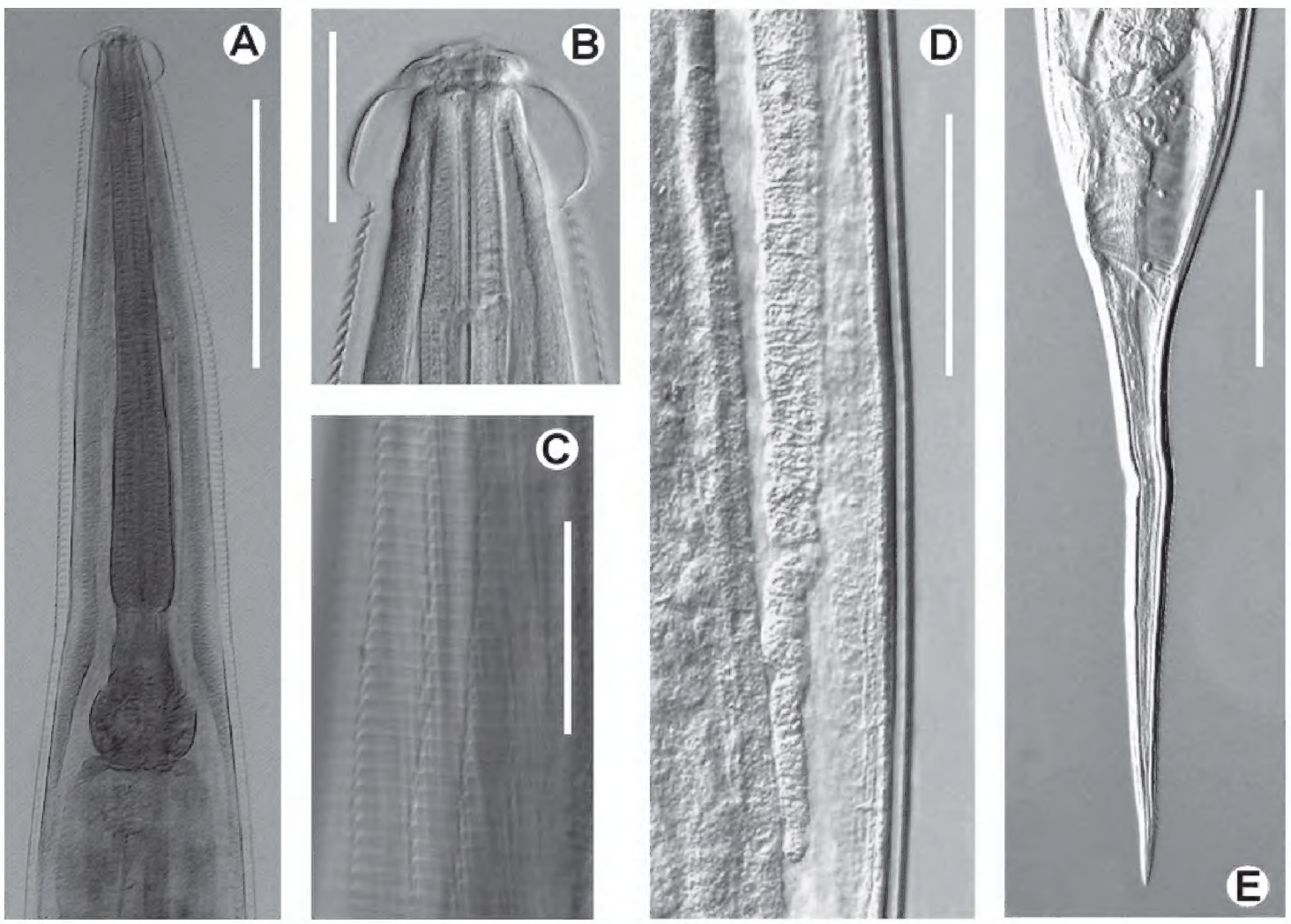

Figure 2. *Hystrignathus splendidus* sp. n. female. **A** Esophageal region **B** Cephalic end, internal view **C** Last cervical spines **D** Unreflexed tip of the posterior ovary **E** Tail, lateral view. Scale bars: **B–D** 0.05 mm **E** 0.1 mm **A** 0.2 mm.

shorter and not reflexed. Eggs ovoid with eight longitudinal but not very prominent ridges on shell. Tail conical, comparatively short, slightly subulate, ending in fine tip. Male unknown.

Differential diagnosis. H. splendidus sp. n. can be easily differentiated from most species of the genus by the long and inflated first cephalic annule. It shares this feature with *H. tarda* (Artigas, 1928) and *H. inflatus* Travassos & Kloss, 1957 from Brazil.

It is close to *H. inflatus* but differs by its ridged eggs which are shorter in length but just as wide (0.098–0.108×0.043–0.053: 0.110–0.129×0.038–0.053). Also, *H. splendidus* sp. n. has a comparatively stout body (a = 14.71–15.89: 19.75–20.07) and comparatively short tail (c = 5.71–7.24: 4.74–5.04).

H. splendidus sp. n. differs from *H. tarda* by the extension of the cervical spines, which in the latter end at the middle of the basal bulb. In *H. tarda* the eggs are smooth and its measurements notably smaller (0.085×0.030 *vs.* 0.098–0.108×0.043–0.053).

Type host. Passalus interstitialis Escholtz, 1829 (Coleoptera: Passalidae).

Site. Gut caeca.

Type locality. Escaleras de Jaruco, Jaruco, La Habana Province, Cuba.

Etymology. Specific epithet is derived from the Latin *splendidus,* splendid, due to the beauty of the species.

Genus *Lepidonema* Cobb, 1898

Key to species of *Lepidonema* Cobb, 1898 from Cuba

1. Body large (>2 mm); spines ending in middle of bulb or short distance posterior to it; lateral alae commencing at distance from end of spines ..... ***L. magnum* Morffe & García, sp. n.**
– Body shorter (<2 mm); spines ending in middle of isthmus; lateral alae commencing at end of spines..... ***L. teresae* García, Ventosa & Morffe, 2009**

Lepidonema magnum sp. n.

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Figs 3 A–I, 4 A–G

Type material. ♀ holotype, Cuba, La Habana Province, Jaruco, Escaleras de Jaruco; in *Passalus interstitialis*; 16.III.2008; E. Fonseca, J. Morffe & F. Alvarez coll.; CZACC 11.4533. 2 ♀♀ paratypes, same data as holotype, CZACC 11.4534, 11.4594.

Measurements. Holotype female a = 14.45, b = 4.93, c = 6.82, V% = 51.66, total length = 2.710, maximum body width = 0.188, first cephalic annule = 0.018×0.083, stoma length = 0.030, procorpus length = 0.435, isthmus length = 0.035, diameter of basal bulb = 0.095, total length of esophagus = 0.550, nerve ring to anterior end = 0.250, excretory pore to anterior end = 0.730, vulva to posterior end = 1.310, anus to posterior end = 0.398, eggs = 0.088–0.095×0.035–0.040 (0.092 ± 0.004×0.037 ± 0.003) (n = 3).

Paratypes (females) (n = 2) a = 13.15–15.09 (14.12 ± 1.37), b = 4.60–4.96 (4.78 ± 0.26), c = 6.90–6.92 (6.91 ± 0.02), V% = 49.81–53.42 (51.61 ± 2.55), total length = 2.415–2.630 (2.523 ± 0.152), maximum body width = 0.160–0.200 (0.180 ± 0.028), first cephalic annule = 0.018×0.075–0.083 (0.018×0.079 ± 0.005), stoma length = 0.030–0.035 (0.033 ± 0.004), procorpus length = 0.400–0.428 (0.414 ± 0.019), isthmus length = 0.030–0.040 (0.035 ± 0.007), diameter of basal bulb = 0.088–0.095 (0.091 ± 0.005), total length of esophagus = 0.525–0.530 (0.528 ± 0.004), nerve ring to anterior end = 0.233–0.245 (0.239 ± 0.009), excretory pore to anterior end = 0.670–0.710 (0.69 ± 0.028), vulva to posterior end = 1.125–1.320 (1.223 ± 0.138), anus to posterior end = 0.350–0.380 (0.365 ± 0.021) eggs = 0.093×0.038 (n = 1).

Description. Female body large and robust. Cuticle markedly annulated in spiny region (annule about 0.008 mm width) and less in rest of body. These annules almost disappear in last third of body. Cervical cuticle with opposite rows of spines from end of first cephalic annule to midpoint of basal bulb or very short distance beyond its end, arranged initially in 16 rows of robust, scale-like spines increasing to about 20 rows of thinner spines where they end. Longitudinal sub-cuticular striae present. Lateral alae well developed, from short distance posterior to end of spines (about 0.2 body-widths) to level of vulva. Head short, bearing eight small papillae arranged in pairs. First ce-

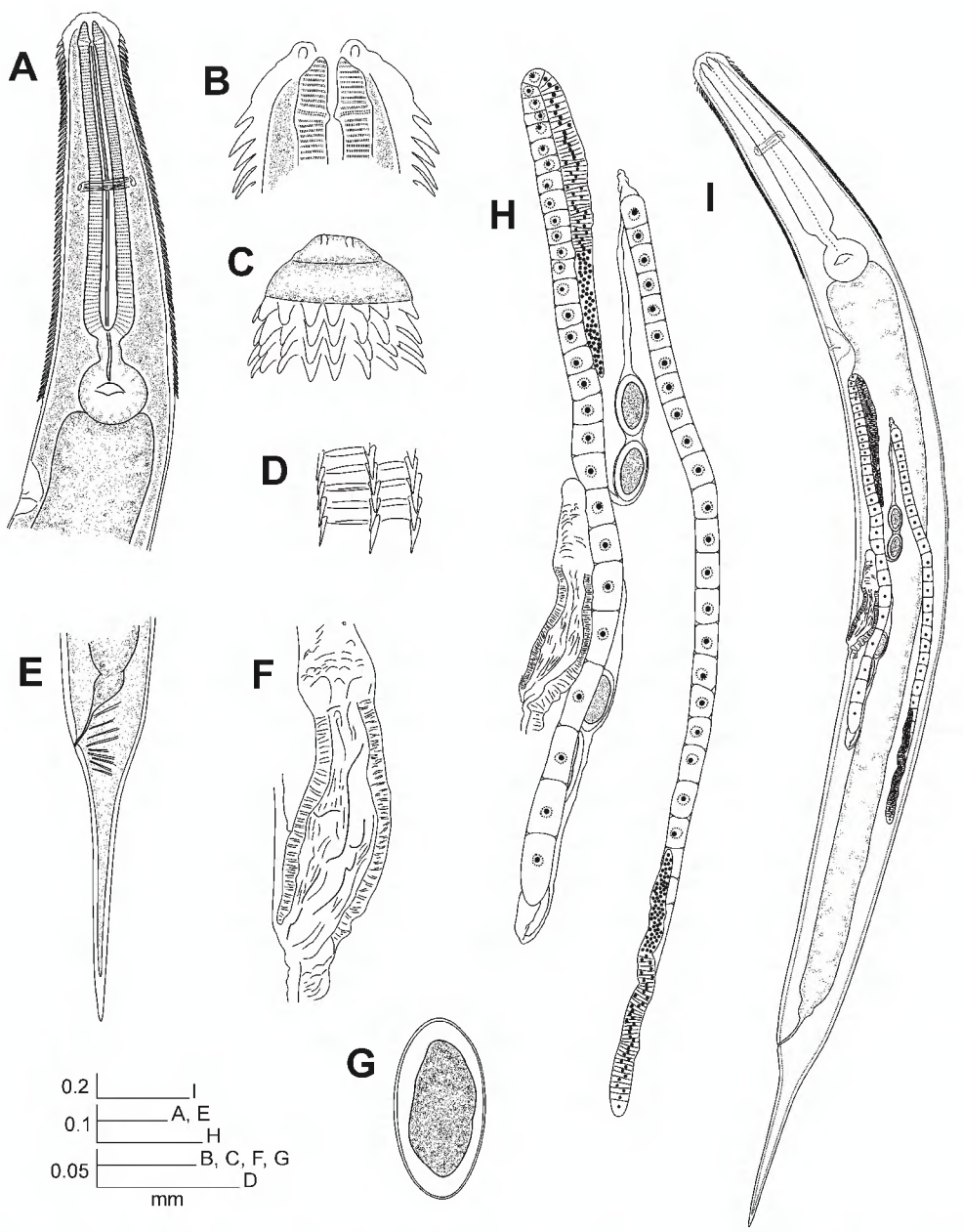

Figure 3. *Lepidonema magnum* sp. n. female. **A** Esophageal region, lateral view **B** Cephalic end, internal view **C** Cephalic end, external view **D** Spines at level of basal bulb **E** Tail, lateral view **F** Vulva, lateral view **G** Egg **H** Genital tract **I** Habitus, lateral view.

phalic annule long, with rounded margins and almost equivalent to head-width. Stoma stout, short, hardly extending posterior to first cephalic annule. Esophagus with powerful, muscular and sub-cylindrical procorpus which has base set off from short isthmus. Basal bulb sub-spherical; valve plate well developed. Intestine simple, sub-rectilinear, with fore region strongly dilated. Rectum short; anus not prominent. Nerve ring surrounding procorpus at about half of its length. Excretory pore situated at little less than body width posterior to basal bulb. Vulva a median transverse slit near mid-body; lips not prominent. Vagina muscular, forwardly directed. Genital tract didelphic-amphidelphic. Ovaries similar in length and reflexed; distal flexures of about 1.5 body-widths in length. Anterior ovary reflexed just posterior to excretory pore. Posterior ovary reflexed at about 2.6 body-widths anterior to anus. Eggs comparatively small, ovoid; shell smooth and thin. Tail short, conical, subulate, ending in fine tip. Male unknown.

Differential diagnosis. L. magnum sp. n. is similar to *L. brasiliensis* Travassos & Kloss, 1957 in body length (2.415–2.710 *vs.* 2.640–2.850), the comparative length of the esophagus (b = 4.60–4.96: 4.71–4.83), spines which cease a short distance pos-

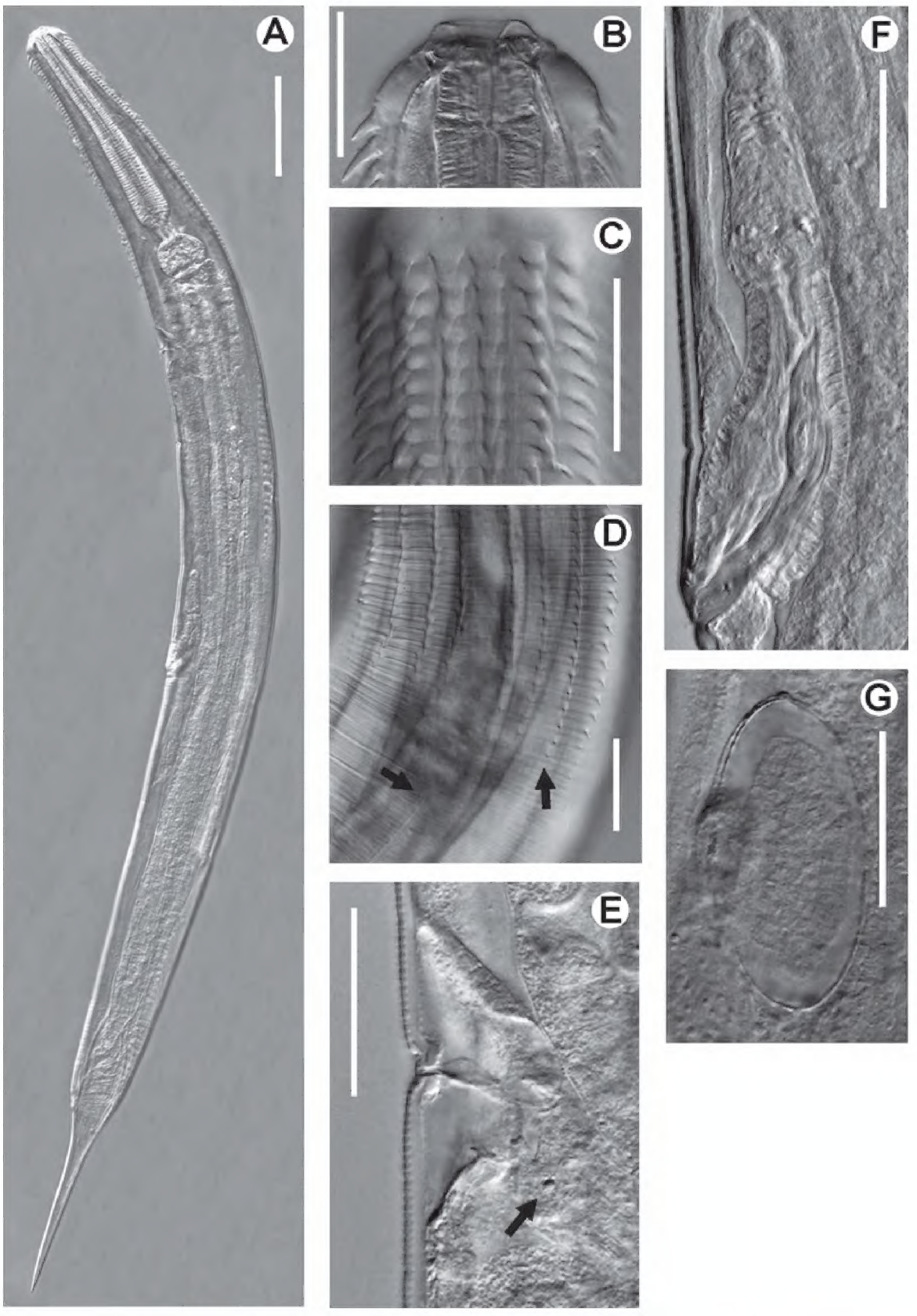

Figure 4. *Lepidonema magnum* sp. n. female. **A** Habitus, lateral view **B** Cephalic end **C** Spines from the cephalic end **D** Last spines and commence of lateral alae (arrows show the end of spines and the commence of lateral alae, respectively) **E** Excretory pore (arrow shows the nuclei of the excretory cell) **F** Vulva, lateral view **G** Egg. Scale bars: **B–G** 0.05 mm **A** 0.1 mm.

terior to the bulb in one specimen (in the other specimens they end at the midpoint of the basal bulb) and the termination of the lateral alae at the level of the vulva. *L. magnum* sp. n. can be distinguished from *L. brasiliensis* by the lateral alae arising at about 0.2 body-widths posterior to the end of the spines but without actually reaching them. In *L. brasiliensis* the lateral alae commence at the end of the spines. Also, *L. magnum* sp. n. has a shorter first cephalic annule (0.018 *vs.* 0.028–0.038), stouter body

(a = 13.15–14.45 *vs.* 17.6–17.81) and the tail is comparatively shorter (c = 6.82–6.92 *vs.* 6.29–6.33).

L. magnum differs from *L. teresae* García, Ventosa & Morffe, 2009 (the only other species of the genus known to occur in Cuba) by its larger body (2.630–2.710 *vs.* 1.610–1.790) and both the esophagus (b = 4.93–4.96: 3.65) and the tail (c = 6.82–6.92 *vs.* 4.10–4.42) are comparatively shorter. As in *L. brasiliensis* the lateral alae of *L. teresae* commence at the end of the spines (García et al. 2009b).

L. bifurcata Cobb, 1898, from Australia, has a bifid tail tip, a unique feature in the genus that differentiates it from the new species. From *L. caracae* Kloss, 1962 in Brazil, it differs by the extension of the lateral alae which, in the latter, end at the level of the anus. *L. caracae* has comparatively shorter esophagus (b = 6.43 *vs.* 4.93–4.96) and the tail (c = 11.59 *vs.* 6.82–6.92). Also, the vulva is more posterior in the Brazilian taxon (V% = 58.96 *vs.* 49.81–53.42).

Type host. Passalus interstitialis Escholtz, 1829 (Coleoptera: Passalidae)

Site. Gut caeca

Type locality. Escaleras de Jaruco, Jaruco, La Habana Province, Cuba.

Etymology. Named after the Latin *magnus*, great or powerful, due to the size and robustness of the species.

Genus *Longior* Travassos & Kloss, 1958

Longior similis Morffe, García & Ventosa, 2009

Material examined 3 ♀♀. Cuba, Matanzas Province, El Pan de Matanzas; in *Passalus interstitialis*; 13.VIII.2009; J. Morffe coll.; CZACC 11.4535–11.4537.

Measurements. Females (n = 3): a = 17.86–19.29 (18.75 ± 0.78), b = 4.24–4.43 (4.35 ± 0.10), c = 6.08–6.43 (6.25 ± 0.17), V% = 50.00–52.34 (51.17 ± 1.65), total length = 2.500–2.700 (2.625 ± 0.109), maximum body width = 0.140, stoma length = 0.060–0.073 (0.068 ± 0.007), procorpus length = 0.470–0.520 (0.497 ± 0.025), isthmus length = 0.028–0.038 (0.033 ± 0.005), diameter of basal bulb = 0.073–0.088 (0.078 ± 0.008), total length of esophagus = 0.590–0.610 (0.603 ± 0.012), nerve ring to anterior end = 0.218–0.225 (0.223 ± 0.004), excretory pore to anterior end = 0.690–0.810 (0.757 ± 0.061), vulva to posterior end = 1.250–1.275 (1.363 ± 0.018), anus to posterior end = 0.400–0.440 (0.420 ± 0.020), eggs = 0.128×0.053 n = 1.

Host. Passalus interstitialis Escholtz, 1829 (Coleoptera: Passalidae).

Site. Gut caeca.

Locality. El Pan de Matanzas, Matanzas Province, Cuba.

Remarks. L. similis was originally described in *P. interstitialis* from Escaleras de Jaruco, Jaruco, La Habana Province, Cuba (Morffe et al. 2009). The El Pan de Matanzas population was obtained from the same host. Both localities belong to the "Alturas de Habana-Matanzas", a chain of hill that extends along the northern part of La Habana and Matanzas Provinces.

The specimens from El Pan de Matanzas exhibit a slightly shorter body and esophagus. The remaining measurements, including De Man's ratios, agree with the type population (Table 1).

Acknowledgements

We are indebted to our colleagues Elier Fonseca, Gunnary León (Universidad de La Habana), Ormaily Madruga (Museo Nacional de Historia Natural), Rayner Núñez and Mayke Hernández (Instituto de Ecología y Sistemática) and the undergraduate student Facundo Alvarez for their help during the field work. To the inhabitants of La Jaula community (La Habana province) and the workers of the Caunabaco dam (Matanzas province) for their hospitality and support. To Yamir Torres MSc (Instituto de Ecología y Sistemática) and Dr. Alejandro Barro (Universidad de La Habana) for their help with the micrographs. We also thank Dr. Luis F. de Armas from the Instituto de Ecología y Sistemática for his critical review of the manuscript and Dr. Pedro Herrera for the revision of the English manuscript. This work was financed by IDEAWILD and the project "Colecciones Zoológicas, su Conservación y Manejo" (DB-06) of the Ministerio de Ciencia, Tecnología y Medio Ambiente, Cuba.

Table 1. Comparative measurements of females of *Longior similis* Morffe, García & Ventosa, 2009 parasites of *Passalus interstitialis* Escholtz, 1829 from the type locality Escaleras de Jaruco, Jaruco, La Habana province, Cuba and El Pan de Matanzas, Matanzas province, Cuba.

Measurements	Escaleras de Jaruco (type locality) n = 9	El Pan de Matanzas n = 3
Total length	2.675-3.075	2.500-2.700
Width	0.120-0.160	0.140
Stoma length	0.058-0.065	0.060-0.073
Procorpus length	0.500-0.570	0.470-0.520
Isthmus length	0.030-0.045	0.028-0.038
Basal bulb diameter	0.068-0.075	0.073-0.088
Esophagus length	0.600-0.680	0.590-0.610
Nerve ring-head	0.213-0.238	0.218-0.225
Excretory pore-head	0.760-0.850	0.690-0.810
Vulva-posterior end	1.250-1.500	1.250-1.275
Anus-posterior end	0.410-0.470	0.400-0.440
Eggs	0.120-0.135×0.043-0.063 n = 18	0.128×0.053 n = 1
a	16.88-22.50	17.86-19.29
b	4.18-4.73	4.24-4.43
c	6.22-6.99	6.08-6.43
V%	51.22-53.70	50.00-52.34

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